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Branch Pattern for Anterior Proboscis Nerves in *Macracanthorhynchus hirudinaceus* (Acanthocephala)

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ABSTRACT: The anterior proboscis nerves of *Macracanthorhynchus hirudinaceus* are associated with the sensory support cell duct and sensory nerves. The entire complex of duct and nerves is largely surrounded by the proboscis retractor muscles. The pair of anterior proboscis nerves branch immediately posterior to the apical sensory organ. Transmission electron microscopy clearly shows this division as well as the morphology of adjacent sensory nerves and sensory support cell duct. The separation of the anterior proboscis nerves into 2 pairs at the base of the apical sensory organ is shown.

KEY WORDS: Acanthocephala, nerves, *Macracanthorhynchus hirudinaceus,* morphology.

Dunagan and Miller (1983) illustrated the relationship of sensory support cell to apical sense organ. Included in this illustration was a drawing showing the entry of the sensory nerves and sensory support cell duct into the cone-shaped apical sense organ. Dunagan and Miller (1983) reported that the anterior proboscis nerves branched at the posterior margin of this organ. Because those observations were made using light microscopy, it was not possible to capture the point where branching occurred. Indeed, the visual evidence presented by these authors requires a bit of faith to interpret. The present study describes observations with transmission electron microscopy (TEM). The greater magnification and thinner sections allow the branching of the anterior proboscis nerves to be shown clearly. In addition, the appearance and initial relationship of the posterior margin of the apical sense organ to the nerves and duct in this region are evident.

Materials and Methods

Macracanthorhynchus hirudinaceus were obtained from pigs through the courtesy of Reelfoot Meat Packaging in Union City, Tennessee. Worms were transported to the laboratory in intestinal contents placed in Dewar flasks. Four to 6 hr elapsed between collection and initial preparation for TEM. The protruded proboscis was removed immediately posterior to the neck and fixed for 1 hr at room temperature in a mixture of 2% glutaraldehyde and 2% freshly prepared formaldehyde in 0.1 M cacodylate buffer (pH 7.2) containing 2.0 mM ethylene glycol-bis (β-aminoethyl ether) N,N,N′,N′-tetraacetic acid (EGTA) and 1.0 mM $MgSO_4$. This preparation was placed in a refrigerator (5°C) for 3 days. Additional procedures were as described by Dunagan and Bozzola (1989). Sections were mounted on slot grids and examined in a Hitachi H500H transmission electron microscope. Position of dorsal and ventral surface is based on conclusions of Hyman (1951).

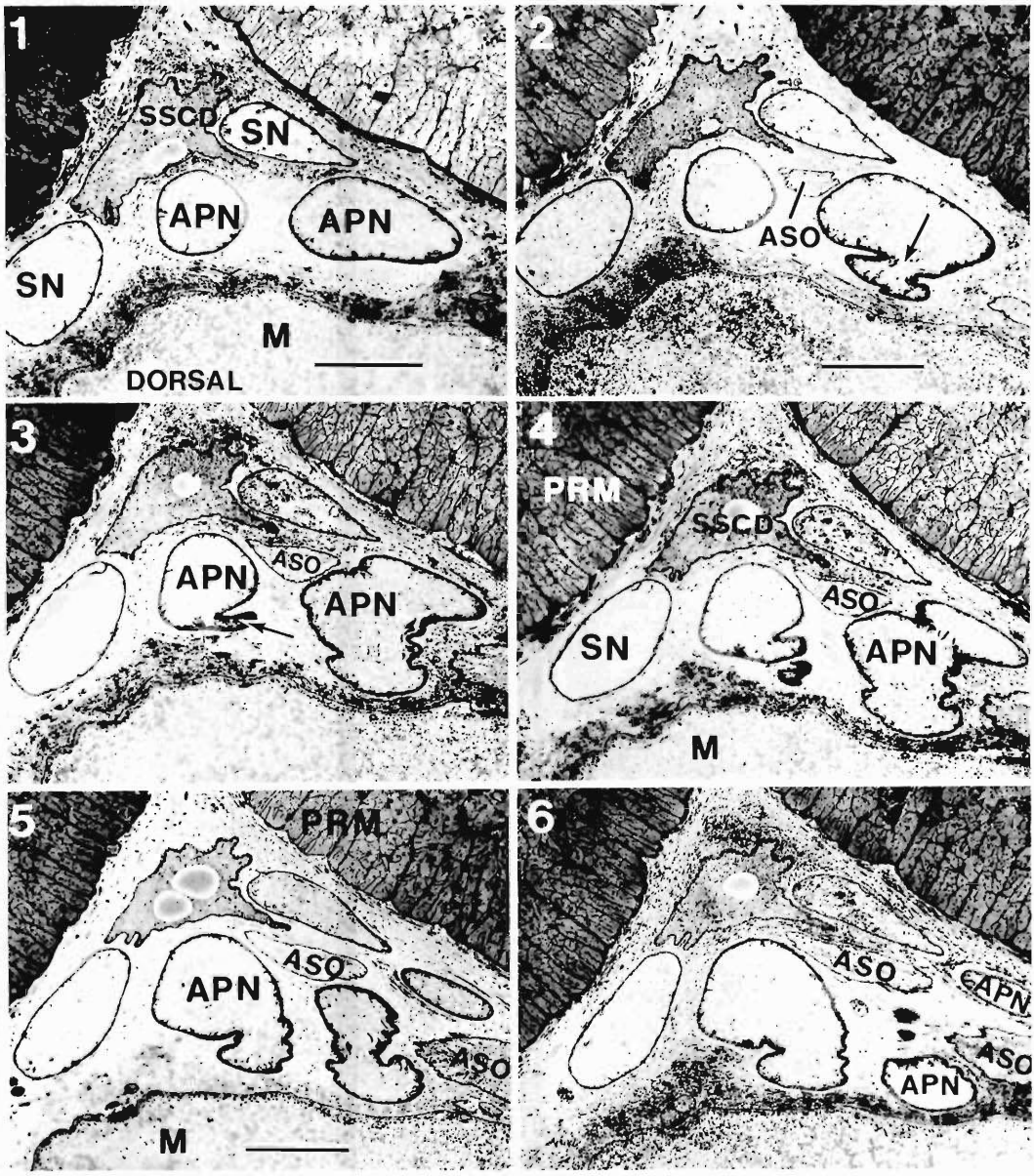
Results

The sensory support cell duct (SSCD), 2 sensory nerves (SN), and 2 anterior proboscis nerves (APN) occupy the same area immediately posterior to the base of the apical sense organ (Fig. 1). These structures are surrounded on 3 sides by proboscis retractor muscles (PRM) and on the fourth by a poorly defined structure called a "Markbeutel" (M) by 19th-century German investigators. At this level in the proboscis, the SSCD outline is very irregular but clearly separate from the SN. More anteriorly the SSCD and SN become so intertwined that they are difficult to separate.

A portion of the apical sense organ (ASO) appears in Figure 2 and gradually expands anteriorly (Figs. 3–10) until it separates the recently branched APN (Fig. 10). The ventral pair of APN will accompany the SSCD and SN until the latter enter the ASO after which both dorsal and ventral APN remain on the outer surface of the ASO.

The two APN do not divide simultaneously. The first division was initiated by the most peripheral APN (Fig. 2, arrow) and progresses (Figs. 2–5) until completed (Fig. 5). The remaining APN begins division more anteriorly (Fig. 3, arrow) and completes division (Fig. 7) after the more peripheral APN is completely separated by the base of the ASO. Following division of the original pair of APN, the newly formed branches and the SN have the same general appearance.

The SSCD remains pleomorphic throughout its remaining length, but structurally it becomes

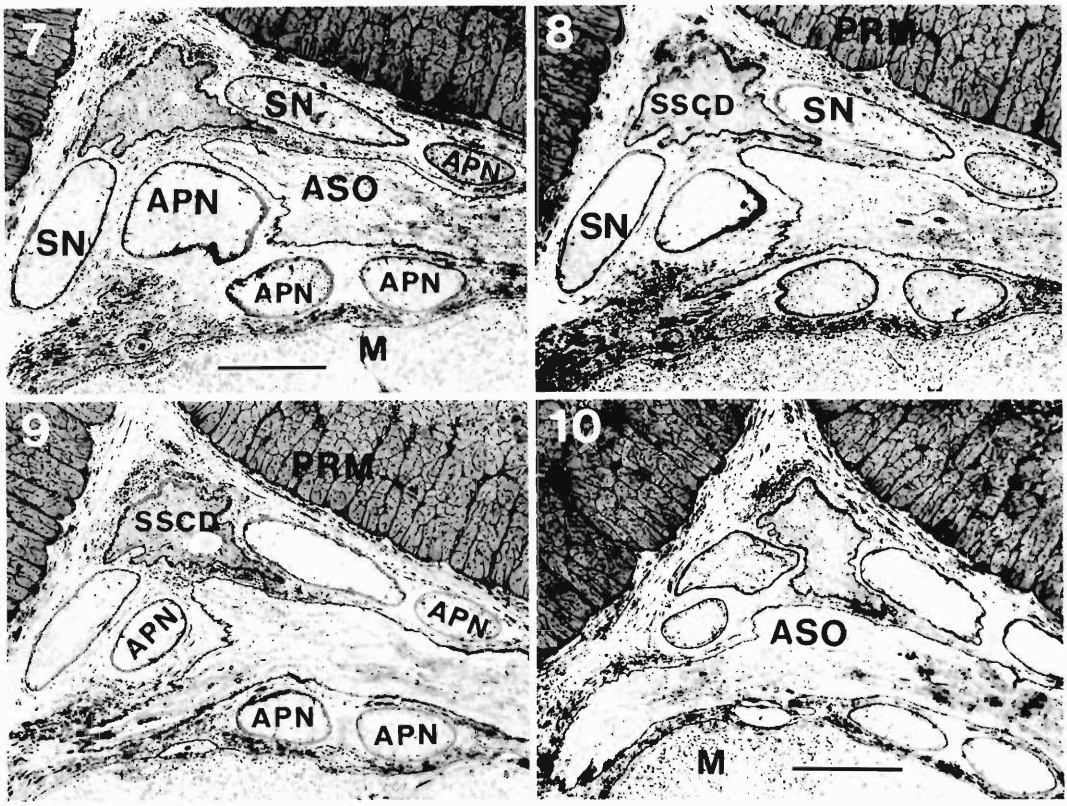


Figures 1–6. Electron photomicrographs of nerves and adjacent tissue in the proboscis of *Macracanthorhynchus hirudinaceus*. 1. Posterior to apical sensory organ. Sensory support cell duct highly vesiculated. 2. Posterior terminus of apical sense organ. One anterior proboscis nerve begins division. 3. Second APN initiates division. 4. Division of first APN nears completion. 5. Division of first APN completed. 6. Division of second APN nears completion. APN, anterior proboscis nerve; ASO, apical sensory organ; M, Markbeutel; PRM, proboscis retractor muscle; SN, sensory nerve; SSCD, sense support cell duct. Magnification ×9,000. Scale bar = 2.0 μm.

increasingly more complex as it approaches the anterior terminus of the ASO. The SN also become pleomorphic and eventually interact with the SSCD. In contrast, the APN are smooth surfaced before and after division. The target of the APN is thought to be retractor muscles.

Discussion

A literature review regarding the general construction of the ASO was made by Dunagan and Miller (1983). This review pointed out the lack of information regarding acanthocephalan recep-



Figures 7–10. Electron photomicrographs of posterior terminus of apical sense organ (ASO) and associated nerves and sensory support cell duct (SSCD) in proboscis of *Macracanthorhynchus hirudinaceus*. 7. Division of second anterior proboscis nerve (APN) completed. 8. Base of ASO separates both recently divided APN. 9. Base of ASO expanded but not yet isolating sensory nerve. 10. Base of ASO completely separates pair of branched APN from SN and SSCD. APN, anterior proboscis nerve; ASO, apical sense organ; M, Markbeutel; PRM, proboscis retractor muscle; SN, sensory nerve; SSCD, sensory support cell duct. Magnification ×9,000. Scale bar = 2.0 μm.

tor morphology as well as the absence of data on receptor function. These authors also reviewed the work of Kaiser (1893), Rauther (1930), and Kilian (1932), all of whom mentioned the presence of nerves at the "Tastpapille." Earlier work in this area was reviewed by Harada (1931). None of these studies was as well received as the study of Brandes (1899) who had his illustration appear in reviews by Hyman (1951) and Bullock and Horridge (1965). Brandes illustrated a single nerve to the ASO. This nerve was unbranched. Dunagan and Miller (1983) described a pair of sensory and anterior proboscis nerves to the ASO. The present investigation shows the point of division of the APN and the subsequent separation of these nerves by the ASO. Moreover, the relationship of this division to the ASO, SSCD, and SN is clearly illustrated in the figures presented herein.

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